Claims

1. A compound of formula (1)

$$R_1–A–X–CH_2–R_3–R_4–L_1 \qquad (1)$$

wherein
A is a group recognized by O^6-alkylguanine-DNA alkyltransferases (AGT) as a substrate;
X is oxygen or sulfur;
R_1 is a group $–R_2–L_2$ or a group R_5;
R_2 and R_4 are, independently of each other, a linker;
R_3 is an aromatic or a heteroaromatic group, or an optionally substituted unsaturated alkyl, cycloalkyl or heterocyclyl group with the double bond connected to CH_2;
R_5 is arylmethyl or heteroarylmethyl or an optionally substituted cycloalkyl, cycloalkenyl or heterocyclyl group;
L_1 is a label, a plurality of same or different labels, a bond connecting R_4 to A forming a cyclic substrate, or a further group $–R_3–CH_2–X–A–R_1$; and
L_2 is a label or a plurality of same or different labels.

2. The compound according to claim 1 of formula (1) wherein
A is a heteroaromatic group containing 1 to 5 nitrogen atoms;
X is oxygen;
R_1 is a group $–R_2–L_2$ or a group R_5;

R_2 and R_4 are, independently of each other, a straight or branched chain alkylene group with 1 to 300 carbon atoms, wherein optionally
(a) one or more carbon atoms are replaced by oxygen, in particular wherein every third carbon atom is replaced by oxygen, e.g. a poylethyleneoxy group with 1 to 100 ethyleneoxy units;
(b) one or more carbon atoms are replaced by nitrogen carrying a hydrogen atom, and the adjacent carbon atoms are substituted by oxo, representing an amide function –NH–CO–;
(c) one or more carbon atoms are replaced by oxygen, and the adjacent carbon atoms are substituted by oxo, representing an ester function –O–CO–;

(d) the bond between two adjacent carbon atoms is a double or a triple bond, representing a function –CH=CH– or –C□C–;
(e) one or more carbon atoms are replaced by a phenylene, a saturated or unsaturated cycloalkylene, a saturated or unsaturated bicycloalkylene, a bridging heteroaromatic or a bridging saturated or unsaturated heterocyclyl group;
(f) two adjacent carbon atoms are replaced by a disulfide linkage –S–S–;
or a combination of two or more, especially two or three, alkylene and/or modified alkylene groups as defined under (a) to (f) hereinbefore, optionally containing substituents;

R_3 is phenyl, an unsubstituted or substituted mono- or bicyclic heteroaryl group of 5 or 6 rings atoms comprising zero, one, two, three or four ring nitrogen atoms and zero or one oxygen atom and zero or one sulfur atom, with the proviso that at least one ring carbon atom is replaced by a nitrogen, oxygen or sulfur atom, 1-alkenyl, 1-alkinyl, 1-cyclohexenyl with 3 to 7 carbon atoms, or an optionally substituted unsaturated heterocyclyl group with 3 to 12 atoms and 1 to 5 heteroatoms selected from nitrogen, oxygen and sulfur, and a double bond in the position connecting the heterocyclyl group to methylene CH_2;

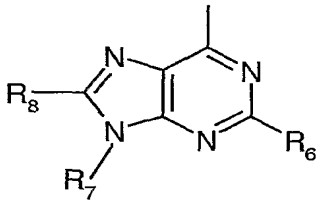
R_5 is optionally substituted phenylmethyl or naphthylmethyl; optionally substituted heteroarylmethyl wherein heteroaryl is a mono- or bicyclic heteroaryl group comprising zero, one, two, three or four ring nitrogen atoms and zero or one oxygen atom and zero or one sulfur atom, with the proviso that at least one ring carbon atom is replaced by a nitrogen, oxygen or sulfur atom, and which has 5 to 12 ring atoms; optionally substituted cycloalkyl with 3 to 7 carbon atoms; optionally substituted cycloalkenyl with 5 to 7 carbon atoms; optionally substituted saturated or unsaturated heterocyclyl with 3 to 12 atoms, and 1 to 5 heteroatoms selected from nitrogen, oxygen and sulfur;

L_1 is one or a plurality of same or different labels selected from a spectroscopic probe, a magnetic probe, a contrast reagent, a molecule which is one part of a specific binding pair which is capable of specifically binding to a partner, a molecule that is suspected to interact with other biomolecules, a library of molecules that are suspected to interact with other biomolecules, a molecule which is capable of crosslinking to other molecules, a molecule which is capable of generating hydroxyl radicals upon exposure to H_2O_2 and

ascorbate, a molecule which is capable of generating reactive radicals upon irradiation with light, a molecule covalently attached to a solid support, a nucleic acid or a derivative thereof capable of undergoing base-pairing with its complementary strand, a lipid or other hydrophobic molecule with membrane-inserting properties, a biomolecule with desirable enzymatic, chemical or physical properties, a bond connecting R_4 to A forming a cyclic substrate, and a further group $-R_3-CH_2-X-A-R_1$; and

L_2 is one or a plurality of same or different labels selected from a spectroscopic probe, a magnetic probe, a contrast reagent, a molecule which is one part of a specific binding pair which is capable of specifically binding to a partner, a molecule that is suspected to interact with other biomolecules, a library of molecules that are suspected to interact with other biomolecules, a molecule which is capable of crosslinking to other molecules, a molecule which is capable of generating hydroxyl radicals upon exposure to H_2O_2 and ascorbate, a molecule which is capable of generating reactive radicals upon irradiation with light, a molecule covalently attached to a solid support, a lipid or other hydrophobic molecule with membrane-inserting properties, and a biomolecule with desirable enzymatic, chemical or physical properties.

3. The compound according to claim 1 of formula (1) wherein the group R_1-A is a purine radical of formula (2)



(2)

wherein R_6 is hydrogen, hydroxy or unsubstituted or substituted amino; and one of R_7 and R_8 is R_1 and the other one is hydrogen.

4. The compound according to claim 3 of formula (1) wherein X is oxygen and R_3 is phenyl.

5. The compound according to claim 3 of formula (1) wherein X is oxygen and R_3 is thienyl.

6. The compound according to claim 3 of formula (1) wherein the group R_1–A is a purine radical of formula (2), R_6 is unsubstituted amino, R_7 is R_1, R_8 is hydrogen, and X is oxygen.

7. The compound according to claim 3 of formula (1) wherein the group R_1–A is a purine radical of formula (2), R_6 is unsubstituted amino, R_7 is a group –R_2–L_2, R_8 is hydrogen, and X is oxygen.

8. The compound according to claim 7 wherein L_2 is a spectroscopic probe.

9. The compound according to claim 7 wherein L_1 and L_2 are spectroscopic probes.

10. The compound according to claim 9 wherein L_1 and L_2 represent a fluorescence donor / fluorescence quencher pair.

11. The compound according to claim 10 wherein L_1 and L_2 represent a FRET pair.

12. The compound according to claim 3 of formula (1) wherein the group R_1–A is a purine radical of formula (2), R_6 is unsubstituted amino, R_7 is a group R_5, R_8 is hydrogen, and X is oxygen.

13. The compound according to claim 12 wherein R_5 is cyclopentyl.

14. The compound according to claim 3 of formula (1) wherein the group R_1–A is a purine radical of formula (2), R_6 is unsubstituted amino, R_7 is hydrogen, R_8 is R_1, and X is oxygen.

15. The compound according to claim 3 of formula (1) wherein the group R_1–A is a purine radical of formula (2), R_6 is unsubstituted amino, R_7 is hydrogen, R_8 is a group –R_2–L_2, and X is oxygen.

16. The compound according to claim 15 wherein L_2 is a spectroscopic probe.

17. The compound according to claim 15 wherein L_1 and L_2 are spectroscopic probes.

18. The compound according to claim 17 wherein L_1 and L_2 represent a fluorescence donor / fluorescence quencher pair.

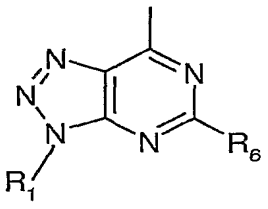
19. The compound according to claim 18 wherein L_1 and L_2 represent a FRET pair.

20. The compound according to claim 15 wherein L_2 is a molecule representing one part of a specific binding pair.

21. The compound according to claim 15 wherein L_2 is a molecule covalently attached to a solid support.

22. The compound according to claim 15 wherein L_2 is a cell membrane transport enhancer group.

23. The compound according to claim 1 of formula (1) wherein the group R_1–A is an 8-azapurine radical of formula (3)



(3)

wherein the substituent R_6 is hydrogen, hydroxy or unsubstituted or substituted amino.

24. The compound according to claim 23 of formula (1) wherein X is oxygen and R_3 is phenyl.

25. The compound according to claim 23 of formula (1) wherein the group R_1–A is an 8-azapurine radical of formula (3), R_6 is unsubstituted amino, R_1 is a group $-R_2-L_2$, and X is oxygen.

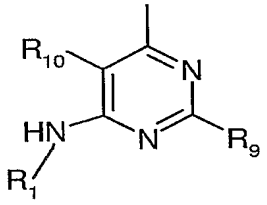
26. The compound according to claim 25 wherein L_2 is a spectroscopic probe.

27. The compound according to claim 25 wherein L_1 and L_2 are spectroscopic probes.

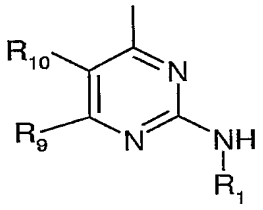
28. The compound according to claim 27 wherein L_1 and L_2 represent a fluorescence donor / fluorescence quencher pair.

29. The compound according to claim 28 wherein L_1 and L_2 represent a FRET pair.

30. The compound according to claim 1 of formula (1) wherein the group R_1–A is a pyrimidine radical of formula (4a) or (4b)



(4a)



(4b)

wherein R_9 is hydrogen, halogen, lower alkyl with 1 to 4 carbon atom or amino, and R_{10} is hydrogen, halogen, lower alkyl with 1 to 4 carbon atoms, amino, nitro or nitroso.

31. The compound according to claim 30 of formula (1) wherein X is oxygen and R_3 is phenyl.

32. The compound according to claim 30 of formula (1) wherein the group R_1–A is a pyrimidine radical of formula (4a) or (4b), R_1 is a group $-R_2-L_2$, and X is oxygen.

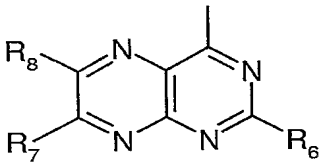
33. The compound according to claim 32 wherein L_2 is a spectroscopic probe.

34. The compound according to claim 32 wherein L_1 and L_2 are spectroscopic probes.

35. The compound according to claim 34 wherein L_1 and L_2 represent a fluorescence donor / fluorescence quencher pair.

36. The compound according to claim 35 wherein L_1 and L_2 represent a FRET pair.

37. The compound according to claim 1 of formula (1) wherein the group R_1–A is a pteridine radical of formula (4c)



(4c)

wherein R_6 is unsubstituted or substituted amino; and one of R_7 and R_8 is R_1 and the other one is hydrogen.

38. The compound according to claim 37 of formula (1) wherein X is oxygen and R_3 is phenyl.

39. The compound according to claim 37 of formula (1) wherein the group R_1–A is a pteridine radical of formula (4c), R_6 is unsubstituted amino, R_7 is hydrogen, R_8 is R_1, R_1 is a group $-R_2-L_2$, and X is oxygen.

40. The compound according to claim 39 wherein L_2 is a spectroscopic probe.

41. The compound according to claim 39 wherein L_1 and L_2 are spectroscopic probes.

42. The compound according to claim 41 wherein L_1 and L_2 represent a fluorescence donor / fluorescence quencher pair.

43. The compound according to claim 42 wherein L_1 and L_2 represent a FRET pair.

44. A method for detecting and/or manipulating a protein of interest, wherein the protein of interest is incorporated into an AGT fusion protein, the AGT fusion protein is contacted with a compound of formula (1) according to any one of claims 1 to 38, and

the AGT fusion protein is detected and optionally further manipulated using the label L_1 in a system designed for recognising and/or handling the label.

45. The method according to claim 44, wherein in the compound of formula (1) label L_2 is a solid support, and the AGT fusion protein contacted with the compound of formula (1) is separated from the compound of formula (1) by filtration or centrifugation or separation of magnetic beads.

46. The method according to claim 44, wherein in the compound of formula (1) label L_1 is one member and label L_2 the other member of two interacting spectroscopic probes L_1 / L_2, and the AGT fusion protein is detected by fluorescence.

47. The method according to claim 44 for detecting and/or manipulating a protein of interest, wherein the protein of interest is fused with a mutant AGT, the mutant AGT fusion protein is contacted with a mixture of
(a) a compound of formula (1) wherein R_1 is a group R_5 and which is not recognized by the mutant AGT, and
(b) another compound of formula (1) recognized by the mutant AGT fusion protein,
and the mutant AGT fusion protein is detected and optionally further manipulated using the label in a system designed for recognizing and/or handling the label.